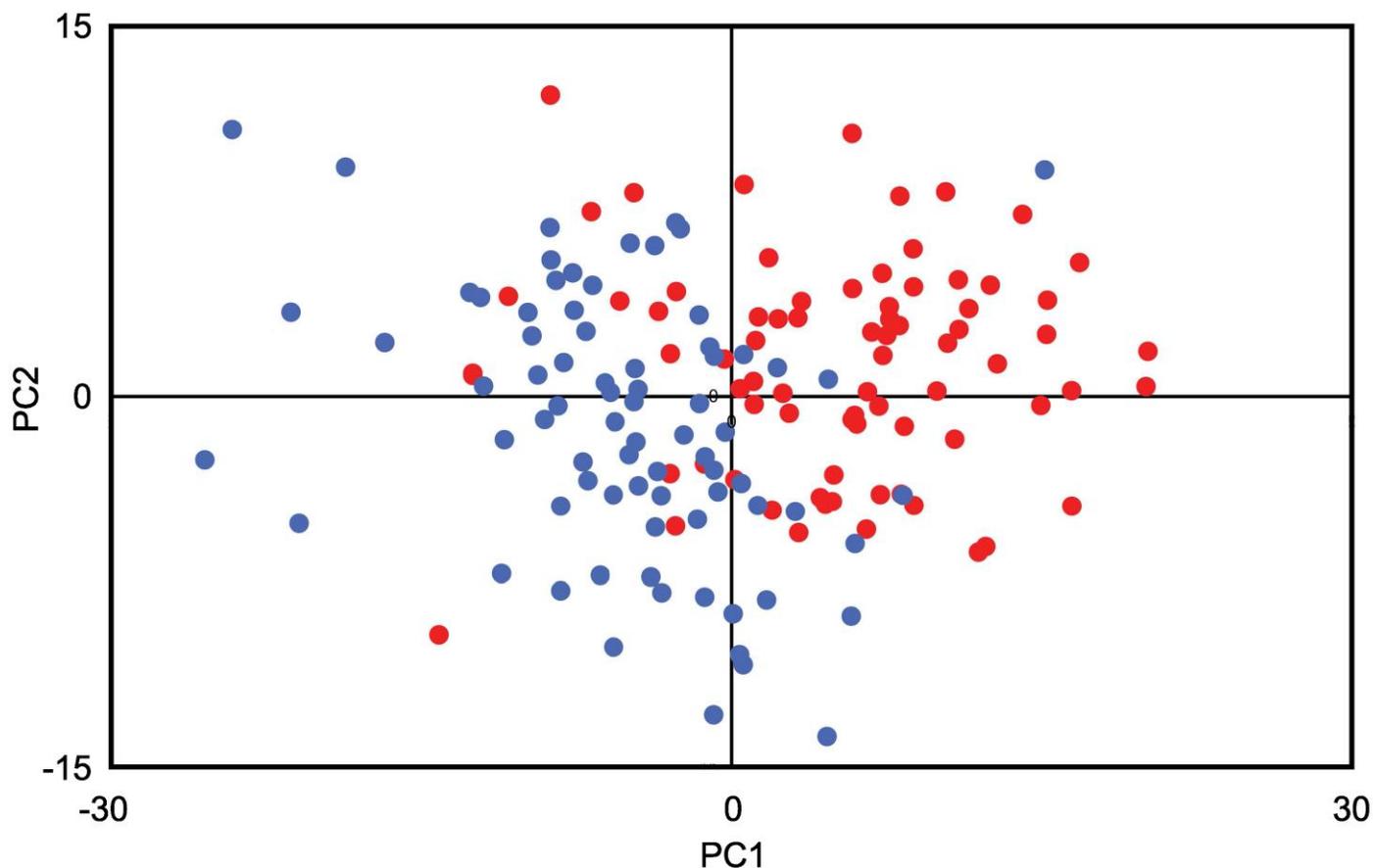
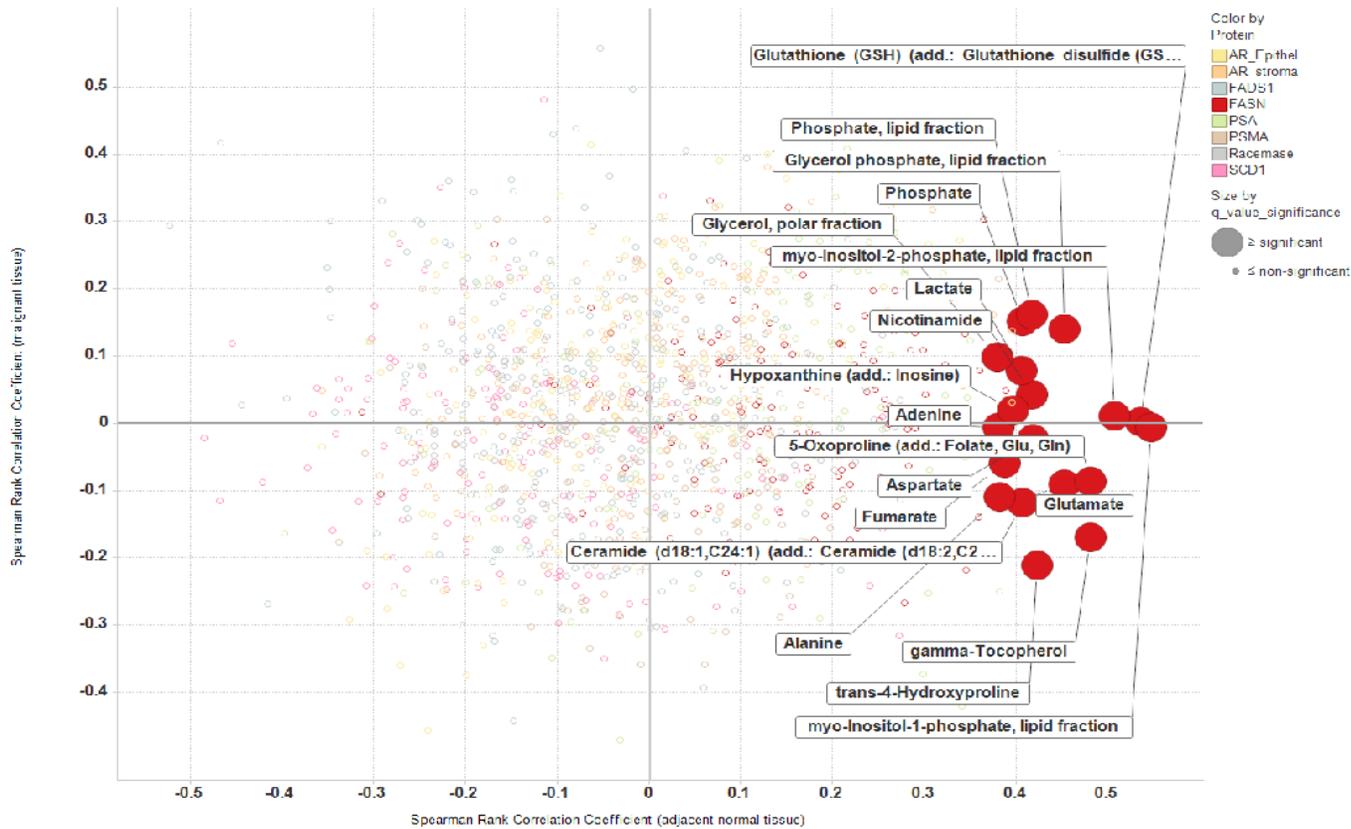


Integration of tissue metabolomics, transcriptomics and immunohistochemistry reveals *ERG*- and gleason score- specific metabolomic alterations in prostate cancer

Supplementary Material



Supplemental Figure 1: Unsupervised analysis of 76 matched malignant and adjacent normal prostate tissue samples. Principal component analysis of subject-corrected residuals (fraction of variance captured by each principal component (PC): PC1: 30.3%; PC2: 9.8%) of MxP® data of malignant samples (red) and adjacent normal samples (blue).



Supplemental Figure 2: Scatter plot of correlation coefficients of tissue-type specific Spearman rank correlation analysis of metabolite ratios versus protein expression data.

Supplemental Table 1: ANOVA results of carcinoma tissue versus adjacent normal as described in the method section. Ratio gives the fold change of carcinoma versus adjacent normal tissue.

Metabolite name	Ontology name	Ratio	p-value	FDR
2-Hydroxybehenic acid (C22:0)	Fatty acids, saturated	4.38	5.34E-15	4.52E-13
Cerebronic acid (2-OH-C24:0)	Fatty acids, hydroxylated	3.49	1.25E-12	3.52E-11
Tricosanoic acid (C23:0)	Fatty acids, saturated	2.53	1.04E-15	2.64E-13
Cystine	Amino acids, S-containing	2.42	6.50E-10	8.69E-09
Nicotineamide adenine dinucleotide (NAD)	Redox-carrier and related	2.16	4.80E-05	1.25E-04
Glycerophosphoethanolamine, polar fraction	Phospholipid metabolites	2.07	8.30E-11	1.26E-04
Uridine	Pyrimidine metabolism	1.82	5.40E-14	1.27E-04
alpha-Tocopherol	Tocopherols and related	1.76	3.12E-06	1.28E-04
Myristic acid (C14:0)	Fatty acids, saturated	1.72	6.86E-09	1.29E-04
Eicosenoic acid (C20:cis[11]1)	Fatty acids, mono-unsaturated	1.66	6.64E-07	1.30E-04
Hypoxanthine (additional: Inosine)	Purine metabolism	1.65	1.02E-13	1.31E-04
Eicosadienoic acid (C20:2) No 02	Fatty acids, poly-unsaturated	1.64	4.76E-09	1.32E-04
Dihydrocholesterol	Cholesterol and related	1.61	4.42E-05	1.33E-04
Xanthine	Purine metabolism	1.59	4.65E-07	1.34E-04
Spermidine	Polyamines	1.59	1.55E-06	1.35E-04
Biotin	C1-carriers and related	1.58	2.74E-15	1.36E-04
Glycerol-3-phosphate, polar fraction	Lipid precursors	1.58	1.71E-09	1.37E-04
Palmitoleic acid (C16:cis[9]1)	Fatty acids, mono-unsaturated	1.58	4.38E-07	1.38E-04
Isopentenyl pyrophosphate (IPP)	Mevalonate pathway	1.57	7.80E-11	1.39E-04
myo-Inositol-1-phosphate, lipid fraction (myo-Inositolphospholipids)	Phospholipid metabolites	1.57	2.02E-08	1.40E-04
myo-Inositol-2-phosphate, lipid fraction (myo-Inositolphospholipids)	Phospholipid metabolites	1.56	2.35E-06	1.41E-04
7-Methylguanine	Purine metabolism	1.54	2.20E-14	1.42E-04
Pentadecanol	Fatty alcohols	1.54	2.49E-08	1.43E-04
beta-Carotene	Carotenoids	1.53	3.09E-06	1.44E-04
Fructose	Glycolysis/Gluconeogenesis	1.52	2.98E-06	1.45E-04
Cysteine (additional: Cystine)	Amino acids, S-containing	1.51	4.21E-12	1.46E-04
2-Amino adipic acid	Amino acid metabolites	1.5	8.35E-07	1.47E-04
Glycerol-2-phosphate	Miscellaneous	1.49	4.03E-10	1.48E-04
Heptadecanoic acid (C17:0)	Fatty acids, saturated	1.48	3.27E-09	1.49E-04

Eicosanoic acid (C20:0)	Fatty acids, saturated	1.48	6.24E-07	1.50E-04
erythro-Dihydrosphingosine (d16:0)	Sphingolipids	1.47	2.34E-06	1.51E-04
Aspartate	Amino acids, acidic	1.45	1.42E-10	1.52E-04
N-Acetylneuraminic acid	Aminosugars	1.43	1.95E-05	1.53E-04
4-Hydroxysphinganine (t18:0, Phytosphingosine), total	Sphingolipids	1.43	1.66E-04	1.54E-04
gamma-Tocopherol	Tocopherols and related	1.42	1.52E-07	1.55E-04
Glycerol, lipid fraction	Fatty alcohols	1.39	2.45E-08	1.56E-04
Cholesterol No 02	Cholesterol and related	1.39	3.46E-07	1.57E-04
Behenic acid (C22:0)	Fatty acids, saturated	1.39	5.76E-07	1.58E-04
Oleic acid (C18:cis[9]1)	Fatty acids, mono-unsaturated	1.38	2.28E-06	1.59E-04
Proline	Amino acids, neutral	1.37	3.06E-09	1.60E-04
Glucose, lipid fraction	Glycolipids	1.37	2.72E-04	1.61E-04
Ribose	Nucleobase related saccharides	1.36	7.26E-08	1.62E-04
Ornithine (additional: Arginine, Citrulline)	Urea cycle and related	1.36	2.14E-06	1.63E-04
Histidine	Amino acids, basic	1.36	4.60E-06	1.64E-04
14-Methylhexadecanoic acid	Fatty acids, branched	1.36	1.22E-05	1.65E-04
S-Adenosylmethionine	Methyl cycle	1.35	1.70E-04	1.66E-04
Elaidic acid (C18:trans[9]1)	Fatty acids, mono-unsaturated	1.35	5.56E-03	1.67E-04
Glycine	Amino acids, neutral	1.34	5.82E-12	1.68E-04
Docosapentaenoic acid (C22:cis[7,10,13,16,19]5)	Fatty acids, poly-unsaturated	1.34	2.37E-08	1.69E-04
Linoleic acid (C18:cis[9,12]2)	Fatty acids, poly-unsaturated	1.33	2.90E-08	1.70E-04
Palmitic acid (C16:0)	Fatty acids, saturated	1.33	1.77E-06	1.71E-04
Uracil	Pyrimidine metabolism	1.32	7.97E-14	1.72E-04
Hypoxanthine, lipid fraction	Miscellaneous	1.32	6.03E-06	1.73E-04
Xylitol	Polyols	1.32	6.12E-05	1.74E-04
Methionine	Amino acids, S-containing	1.31	5.49E-09	1.75E-04
Eicosapentaenoic acid (C20:cis[5,8,11,14,17]5)	Fatty acids, poly-unsaturated	1.31	5.98E-04	1.76E-04
trans-4-Hydroxyproline	Collagen metabolism	1.3	3.12E-10	1.77E-04
Fumarate	Citrate cycle	1.3	3.72E-07	1.78E-04
N-Acetylneuraminic acid, lipid fraction	Glycolipids	1.3	9.36E-05	1.79E-04
Isoleucine	Amino acids, branched chain	1.29	1.82E-10	1.80E-04
Pantothenic acid	Acyl-carriers and related	1.29	3.43E-06	1.81E-04
Malate	Citrate cycle	1.29	5.68E-06	1.82E-04
Sphingosine (d18:1) isomer No 03	Sphingolipids	1.29	1.15E-04	1.83E-04
Sphingomyelin (d18:1,C23:0)	Sphingomyelins	1.28	9.18E-14	1.84E-04
Docosahexaenoic acid (C22:cis[4,7,10,13,16,19]6)	Fatty acids, poly-unsaturated	1.28	6.51E-06	1.85E-04
Lignoceric acid (C24:0)	Fatty acids, saturated	1.28	8.68E-06	1.86E-04
3-O-Methylsphingosine (d18:1)	Sphingolipids	1.28	3.43E-05	1.87E-04

(additional: Sphingolipids, erythro-Sphingosine (d18:1), threo-Sphingosine (d18:1))				
erythro-Sphingosine-1-phosphate (d18:1)	Sphingolipids	1.28	2.41E-04	1.88E-04
Leucine	Amino acids, branched chain	1.27	2.58E-08	1.89E-04
Phenylalanine	Amino acids, aromatic	1.27	8.60E-08	1.90E-04
Threonine	Amino acids, neutral	1.27	2.91E-07	1.91E-04
Lysine	Amino acids, basic	1.27	8.75E-06	1.92E-04
Flavine adenine dinucleotide (FAD)	Redox-carrier and related	1.27	1.25E-04	1.93E-04
threo-Sphingosine (d18:1) (additional: Sphingolipids)	Sphingolipids	1.27	4.98E-04	1.94E-04
Glutamate	Amino acids, acidic	1.26	1.22E-08	1.95E-04
Tyrosine	Amino acids, aromatic	1.26	4.30E-06	1.96E-04
Glutamine	Amino acids, basic	1.26	1.19E-05	1.97E-04
5-O-Methylsphingosine (d18:1) (additional: Sphingolipids, erythro-Sphingosine (d18:1), threo-Sphingosine (d18:1))	Sphingolipids	1.26	1.51E-04	1.98E-04
Citrulline	Urea cycle and related	1.25	4.62E-06	1.99E-04
Glycerol phosphate, lipid fraction	Phospholipid metabolites	1.25	1.67E-05	2.00E-04
dihomo-gamma-Linolenic acid (C20:cis[8,11,14]3)	Fatty acids, poly-unsaturated	1.25	5.65E-05	2.01E-04
erythro-Sphingosine (d18:1) (additional: Sphingolipids)	Sphingolipids	1.24	6.61E-05	2.02E-04
Phosphate (inorganic and from organic phosphates)	Miscellaneous	1.23	5.05E-08	2.03E-04
Succinate	Citrate cycle	1.23	2.61E-07	2.04E-04
Stearic acid (C18:0)	Fatty acids, saturated	1.23	7.92E-06	2.05E-04
myo-Inositol, lipid fraction	Glycolipids	1.22	1.87E-03	2.06E-04
5-Oxoproline (additional: Folic acid, Glutamate, Glutamine)	Amino acid metabolites	1.21	5.52E-09	2.07E-04
Tryptophan	Amino acids, aromatic	1.21	3.43E-06	2.08E-04
Lysophosphatidylcholine (C18:2)	Lysophosphatidylcholines	1.21	1.47E-05	2.09E-04
Hypotaurine	Amino acid metabolites	1.21	2.97E-03	2.10E-04
Ribulose-5-phosphate	Pentose phosphate pathway	1.21	6.77E-03	2.11E-04
Homoserine	Amino acid metabolites	1.2	2.04E-06	2.12E-04
Nervonic acid (C24:cis[15]1)	Fatty acids, mono-unsaturated	1.2	8.15E-05	2.13E-04
Serine	Amino acids, neutral	1.2	1.15E-04	2.14E-04
Valine	Amino acids, branched chain	1.19	9.78E-06	2.15E-04
Guanosine	Purine metabolism	1.19	7.74E-05	2.16E-04
Arginine	Amino acids, basic	1.19	1.15E-04	2.17E-04
erythro-Dihydrosphingosine (d18:0)	Sphingolipids	1.19	4.02E-03	2.18E-04
Alanine	Amino acids, neutral	1.18	2.40E-05	2.19E-04

Adenine, lipid fraction	Miscellaneous	1.18	3.44E-02	2.20E-04
Threonic acid	Ascorbic acid and related	1.17	2.59E-03	2.21E-04
Pyrophosphate (PPi) (additional: Phosphate (inorganic and from organic phosphates))	Miscellaneous	1.17	8.61E-03	2.22E-04
Cytosine (additional: 2'-Deoxycytidine)	Pyrimidine metabolism	1.16	1.62E-08	2.23E-04
Pseudouridine	Pyrimidine metabolism	1.16	2.72E-04	2.24E-04
Phosphate, lipid fraction	Phospholipid metabolites	1.15	3.06E-04	2.25E-04
Sarcosine	Methyl cycle	1.15	2.60E-02	2.26E-04
Adenine	Purine metabolism	1.14	6.29E-06	2.27E-04
Glycerol, polar fraction	Lipid precursors	1.13	1.17E-04	2.28E-04
Nicotinamide	Redox-carrier and related	1.13	2.74E-04	2.29E-04
Cholesterol, total	Cholesterol and related	1.13	6.15E-04	2.30E-04
Glutathione (GSH) (additional: Glutathione disulfide (GSSG))	Redox-carrier and related	1.13	1.21E-03	2.31E-04
DAG (C18:1,C18:2)	Diacylglycerols	1.13	9.92E-03	2.32E-04
Pyruvate (additional: Phosphoenolpyruvate (PEP))	Glycolysis/Gluconeogenesis	1.12	4.81E-04	2.33E-04
Serine, lipid fraction	Miscellaneous	1.12	9.57E-03	2.34E-04
Coenzyme Q10	Redox-carrier and related	1.1	4.39E-02	2.35E-04
Lysophosphatidylcholine (C20:4)	Lysophosphatidylcholines	1.08	7.25E-03	2.36E-04
Arachidonic acid (C20:cis[5,8,11,14]4)	Fatty acids, poly-unsaturated	1.07	2.60E-02	2.37E-04
Dodecanol	Fatty alcohols	1.07	4.53E-02	2.38E-04
Ceramide (d18:1,C24:1) (additional: Ceramide (d18:2,C24:0))	Ceramides	1.06	8.55E-03	2.39E-04
2-Hydroxybutyrate	Energy metabolism, miscellaneous	1.05	2.70E-02	2.40E-04
Phosphatidylcholine No 02	Phosphatidylcholines	0.96	2.83E-02	2.41E-04
Creatine	Creatine metabolism	0.93	1.11E-02	2.42E-04
myo-Inositol	Polyols	0.91	4.07E-03	2.43E-04
Phosphatidylcholine (C18:0,C20:4)	Phosphatidylcholines	0.89	3.36E-05	2.44E-04
Phosphatidylcholine (C18:0,C22:6)	Phosphatidylcholines	0.86	2.34E-04	2.45E-04
Glutathione disulfide (GSSG)	Redox-carrier and related	0.85	9.29E-03	2.46E-04
Glucuronic acid	Sugar acids	0.72	1.07E-03	2.47E-04
cis-Aconitate (additional: Citrate)	Citrate cycle	0.71	7.58E-03	2.48E-04
Putrescine (additional: Agmatine)	Polyamines	0.71	9.77E-03	2.49E-04
Glucose	Monosaccharides	0.7	1.68E-03	2.50E-04
Gluconic acid (additional: Gluconolacton)	Sugar acids	0.69	6.87E-04	2.51E-04
Spermine	Polyamines	0.69	6.22E-03	2.52E-04
Maltose	Disaccharides	0.65	4.42E-06	2.53E-04

Maltotriose	Trisaccharides	0.6	1.03E-04	2.54E-04
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Supplemental Table 2: Significantly changed protein expression data of 41 matched malignant and adjacent normal prostate tissue samples. Statistical analysis was done via a two-sided paired Wilcoxon signed rank test, the significance level was set to $p < 0.05$.

Analysis	Protein	Direction of change	p-value of paired Wilcoxon signed rank test
Malignant versus adjacent normal	AR (Epithelium)	increase	0.0260
	AR (Stroma)	decrease	0.0171
	PSA	decrease	< 0.0001
	Racemase	increase	< 0.0001
	FASN	increase	0.0001
	PSMA	increase	0.0009
	SCD1	increase	0.0292
	GSTP1	decrease	< 0.0001